

PROJECT NUMBER: 1902  
PROJECT TITLE: Tobacco Microbiology  
PROJECT LEADER: D. J. Ayers  
WRITTEN BY: D. K. Chadick  
PERIOD COVERED: June, 1988

## I. MICROBIAL ANALYSES OF MOLD

- A. Objective: To determine a more rapid method for the enumeration of mold.
- B. Results: Several studies were conducted which compared the mold counts obtained from Czapek Dox agar vs. HC agar, an agar used in the cosmetic industry for the analysis of mold. According to the literature, the HC agar required only 3 days incubation vs. the current 7 day incubation period using Czapek Dox agar (1). However, the data showed that the mold counts from HC agar after incubation for 3 and 7 days were lower than the mold numbers observed from the Czapek Dox agar after 7 days of incubation (1).
- C. Conclusions: Continue to analyze for mold using the laboratory SOP (2).
- D. Plans: Report results in a memo.
- E. References:
1. Gaines, O. Notebook No. 8505, p. 181-186.
  2. Crockett, E. A. Standard Operating Procedure for the Enumeration of Bacteria and Fungi from Tobacco and Tobacco Sheet. PM Special Report, Accession No. 86-055; 1986 February 20.

## II. SHREDDED STEM STUDY (STORAGE OF POST-ART DRIED SAMPLES)

- A. Objective: To determine the effect of long term storage on microbial counts in the post-ART dried samples (12% OV).
- B. Results: A decrease in bacterial counts was observed after 12 weeks of storage in the post-ART dried samples from studies 1, 2, 3, and 5 compared to the initial numbers. The 12 week bacterial counts from study 4 showed a slight but insignificant increase (1.2 fold) when compared to the initial counts (1). The 12 week mold and yeast counts were within the acceptable range (1).
- C. Conclusions: The study is complete.
- D. Plans: A memo will be issued detailing the results.
- E. References:
1. Gaines, O. Notebook No. 8505, p. 160, 162, 166, 170-173.

2000832161

### III. RCB ALTERNATE HUMECTANT PROGRAM (BL PLANT RUN)

- A. Objective: To determine if RCB (control and test sheets), produced as part of the Alternate Humectant Program, differ in microbial numbers.
- B. Results: After 4 and 8 weeks of storage the RCB control and test sheets exhibited a decrease in mold and bacterial counts compared to the time zero values (1).
- C. Plans: This is an ongoing study.
- D. References:
1. Jones, J. Notebook No. 8590, p. 34-35, 40.

### IV. BACTERIAL IDENTIFICATION

- A. Objective: To identify bacterial isolates from different sources.
- B. Results: The SOP for bacterial identification (1), using the Vitek along with supplemental biochemical tests, has been used to identify eight bacterial isolates from laboratory spoiled SEL. Based on the identification the SEL isolates have been characterized into three groups; six of the cultures were Bacillus coagulans, one isolate was B. sphaericus, and one was B. stearothermophilus. The group of B. coagulans was further separated into three separate strains (2).
- C. Conclusions: The study is complete.
- D. Plans: A memo will be issued detailing the previously mentioned results.
- E. References:
1. Chadick, D. Standard Operating Procedure for the Identification of Bacteria with special emphasis on the genus Bacillus, in preparation.
  2. Chadick, D., PM Notebook 8625, p. 39-38, 59, 61-62.

2000832162